

Claims

1. An isolated polynucleotide comprising a nucleic acid molecule one or more selected from the group consisting of
- (a) nucleic acid molecules encoding at least the mature form of the polypeptide depicted in  
5 SEQ ID NO:3;
- (b) nucleic acid molecules comprising the coding sequence as depicted in SEQ ID NO:2;
- (c) nucleic acid molecules whose nucleotide sequence is degenerate as a result of the genetic code to a nucleotide sequence of (a) or (b);
- (e) nucleic acid molecules encoding a polypeptide derived from the polypeptide whose  
10 sequence has an identity of 51.3 % or more to the amino acid sequence of the polypeptide encoded by a nucleic acid molecule of (a) or (b);
- (g) nucleic acid molecules comprising a polynucleotide having a sequence of a nucleic acid molecule amplified from a *Phaffia* nucleic acid library using the primers depicted in SEQ ID NO:4, 5, and 6;
- (j) nucleic acid molecules encoding a polypeptide having squalene synthase activity,  
15 wherein said polypeptide is recognized by antibodies that have been raised against a polypeptide encoded by a nucleic acid molecule of any one of (a), (b), (c) and (g);
- (k) nucleic acid molecules obtainable by screening an appropriate library under stringent conditions with a probe having the sequence of the nucleic acid molecule of any one of (a),  
20 (b), (c), (e), (g) and (j), and encoding a polypeptide having squalene synthase activity.
2. An isolated polynucleotide comprising a nucleic acid molecule one or more selected from the group consisting of:
- (m) nucleic acid molecules comprising the nucleotide sequence as depicted in SEQ ID NO:1;
- (n) nucleic acid molecules whose nucleotide sequence is degenerate as a result of the  
25 genetic code to a nucleotide sequence of (m);
- (p) nucleic acid molecules encoding a polypeptide derived from the polypeptide whose sequence has an identity of 51.3 % or more to the amino acid sequence of the polypeptide encoded by a nucleic acid molecule of (m);

- (q) nucleic acid molecules comprising a fragment encoded by a nucleic acid molecule of any one of (m), (n) or (p) and having squalene synthase activity;
- (r) nucleic acid molecules comprising a polynucleotide having a sequence of a nucleic acid molecule amplified from a *Phaffia* nucleic acid library using the primers depicted in SEQ
- 5 ID NO:4, 5, and 6;
- (s) nucleic acid molecules encoding a polypeptide having squalene synthase activity, wherein said polypeptide is a fragment of a polypeptide encoded by any one of (m), (n), (p), (q) and (r);
- (t) nucleic acid molecules comprising at least 15 nucleotides of a polynucleotide of (m) or
- 10 (a);
- (u) nucleic acid molecules encoding a polypeptide having squalene synthase activity, wherein said polypeptide is recognized by antibodies that have been raised against a polypeptide encoded by a nucleic acid molecule of any one of (m), (n), (p), (q), (r) and (s);
- 15 (v) nucleic acid molecules obtainable by screening an appropriate library under stringent conditions with a probe having the sequence of the nucleic acid molecule of any one of (m), (n), (p), (q), (r), (s), (t) and (u), and encoding a polypeptide having squalene synthase activity;
- (w) nucleic acid molecules whose complementary strand hybridizes under stringent
- 20 conditions with a nucleic acid molecule of any one of (m), (n), (p), (q), (r), (s), (t), (u), (v), and encoding a polypeptide having squalene synthase activity.
3. The isolated polynucleotide of claim 1 or 2, wherein said polynucleotide encodes an amino acid sequence which is identified by SEQ ID NO:3 or has an identity of 51.3 % or more with SEQ ID NO:3.
- 25 4. The isolated polynucleotide of any one of claims 1 to 3, wherein said polynucleotide is derived from a strain of *Phaffia rhodozyma* or *Xanthophylomyces dendrorhous*.
5. A method for making a recombinant vector comprising inserting the polynucleotide of any one of claims 1 to 4 into a vector.
6. A recombinant vector containing the polynucleotide of any one of claims 1 to 4 or
- 30 produced by the method of claim 5.
7. The vector of claim 6 in which the polynucleotide of any one of claims 1 to 4 is operatively linked to expression control sequences allowing expression in prokaryotic or eukaryotic cells.

8. A method of making a recombinant organism comprising introducing the vector of claim 6 or 7 into a host organism.
9. The method of claim 8, wherein said host organism is selected from *E. coli*, baculovirus, or *S. cerevisiae*.
- 5 10. The recombinant organism containing the vector of claim 6 or 7, or produced by the method of claim 8 or 9.
11. A process for producing a polypeptide having squalene synthase activity comprising culturing the recombinant organism of claim 10 and recovering the polypeptide from the culture of said recombinant organism.
- 10 12. A polypeptide obtainable by the process of claim 11.
13. An antibody that binds specifically to the polypeptide of claim 12.
14. An antisense polynucleotide against the polynucleotide of any one of claims 1 to 4.
15. A method for making a recombinant vector comprising inserting the polynucleotide of claim 14 into a vector.
- 15 16. A recombinant vector containing the polynucleotide of claim 14 or produced by the method of claim 15.
17. The vector of claim 16 in which the polynucleotide of claim 14 is operatively linked to expression control sequences allowing expression in prokaryotic or eukaryotic cells.
18. A method of making a recombinant organism comprising introducing the vector of
- 20 claim 16 or 17 into a host organism.
19. The method of claim 18, wherein said host organism belongs to a strain of *Phaffia rhodozyma* or *Xanthophylomyces dendrorhous*.
20. The recombinant organism containing the vector of claim 16 or 17, or produced by the method of claim 18 or 19.
- 25 21. The recombinant organism of claim 20, wherein said organism is characterized in that whose gene expression of squalene synthase is reduced compared to the host organism, thereby is capable of producing carotenoids in an enhanced level relative to a host organism.

22. The recombinant organism according to claim 21, wherein the gene expression of squalene synthase is reduced by means of the technique selected from antisense technology, site-directed mutagenesis, error prone PCR, or chemical mutagenesis.

23. A process for producing carotenoids, which comprises cultivating the recombinant  
5 organism of claim 21.

24. The process of claim 23, wherein said carotenoids are selected one or more from astaxanthin,  $\beta$ -carotene, lycopene, zeaxanthin, canthaxanthin.

25. The process according to claim 23, wherein the gene expression of squalene synthase is reduced in the recombinant organism of claim 21 by means of the technique selected from  
10 antisense technology, site-directed mutagenesis, error prone PCR, or chemical mutagenesis.

26. A process for the production of a carotenoid by culturing a microorganism under suitable conditions and, optionally, recovering the resulting carotenoid, wherein the microorganism is characterized in that its gene expression of squalene synthase is reduced, e.g.  
15 by means of the technique selected from antisense technology, site-directed mutagenesis, error prone PCR, or chemical mutagenesis.

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